510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY TEMPLATE

A. 510(k) Number:

K052366

B. Purpose for Submission:
Addition of the antibiotic Tigecycline at concentrations of 0.016 - 256 µg/mL to the Etest® in the MIC range of 0.016 – 256 µg/mL with Gram positive and Gram negative aerobic bacteria, *Streptococcus species* other than *S. pneumoniae*, and anaerobic bacteria.

C. Measurand:
Tigecycline at 0.016 – 256 µg/mL

D. Type of Test:

Manual Antimicrobial Susceptibility Test System—growth based

E. Applicant:

AB BIODISK

F. Proprietary and Established Names:

Etest® Antimicrobial Susceptibility Test – Tigecycline at 0.016 – 256 µg/mL

G. Regulatory Information:

1. Regulation section:
   21 CFR 866.1640 – Antimicrobial susceptibility test powder
2. Classification:
   Class II
3. Product Code:
   JWY – Manual Antimicrobial Susceptibility Test Systems
4. Panel:
   83 Microbiology

H. Intended Use:

1. Intended use(s):
   Tigecycline at 0.016 – 256 µg/mL for *in vitro* diagnostic use for MIC determination with Gram positive and Gram negative aerobic bacteria, *Streptococcus* species other than *S. pneumoniae*, and anaerobic bacteria.

Etest® is a quantitative technique for the determination of antimicrobial susceptibility of both non-fastidious Gram negative and Gram positive aerobic bacteria, such as *Enterobacteriaceae, Pseudomonas, Staphylococcus* and *Enterococcus* species and fastidious bacteria, such as anaerobes, *N. gonorrhoeae, S. pneumoniae*, *Streptococcus,* and *Haemophilus* species. The system comprises a predefined antibiotic gradient which is used to determine the Minimum Inhibitory Concentration (MIC) in µg/mL of different antimicrobial agents against microorganisms as tested on agar media by overnight incubation.

2. Indication(s) for use:

This submission is for the addition of antibiotic Tigecycline to the Etest® for MIC determination in the MIC range of 0.016 – 256 µg/mL with Gram positive and Gram negative aerobic bacteria, *Streptococcus* species other than *S. pneumoniae*, and anaerobic bacteria.

3. Special condition for use statement(s):
Prescription Use Only

4. Special instrument Requirements:
Manual readings only

I. Device Description:

The Etest® gradient technology is based on a combination of the concepts of dilution and diffusion test methods for susceptibility testing. Etest® directly quantifies antimicrobial susceptibility in terms of discrete MIC values. When the Etest® strip is applied to an inoculated agar plate, the antibiotic is immediately released from the plastic surface into the agar. A predefined, continuous gradient of antibiotic concentrations is created and maintained directly underneath the strip. After incubation whereby bacterial growth becomes visible, a symmetrical inhibition ellipse centered along the strip will be observed. The MIC value in µg/mL is read where the ellipse edge intersects the strip. Since Etest® generates MIC values which fall between two-fold dilutions for interpretation; the MIC value read must be recorded to the next higher two-fold dilution.

Etest® consists of a thin, inert and non-porous plastic strip, which is 5 mm wide and 60 mm long. Strips are supplied in a plastic blister package with 10 units in each of 10 individually sealed compartments. One side of the strip is labeled with the MIC reading scale in µg/mL and a three-letter code on the handle to designate the identity of the antibiotic. The other side of the strip is impregnated with a predefined, dried and stabilized, exponential gradient of an antibiotic, expressed

as a minimum and a maximum value. The gradient covers a continuous concentration range across 15 two-fold dilutions.

J. Substantial Equivalence Information:

1. Predicate device name(s):
   The E Test®

2. Predicate K number(s):
   K913459

3. Comparison with predicate:

Similarities		
Item	**Device**	**Predicate**
Intended use	Etest® is a quantitative technique for the determination of antimicrobial susceptibility of both non-fastidious Gram negative and Gram positive aerobic bacteria, such as *Enterobacteriaceae, Pseudomonas, Staphylococcus* and *Enterococcus* species and fastidious bacteria, such as anaerobes, *Pneumococcus*, *Streptococcus, Gonococcus* and *Haemophilus* species. The system comprises a predefined antibiotic gradient which is used to determine the Minimum Inhibitory Concentration (MIC) in µg/mL of different antimicrobial agents against microorganisms as tested on agar media by overnight incubation.	same
Isolates	Isolated colonies from culture used	same
Results	Report results as minimum inhibitory concentration (MIC)	same
Type of Test	Manual read only	same

Differences		
Item	**Device**	**Predicate**
Antibiotic	Tigecycline	Various antibiotics

4. Standard/Guidance Document Referenced (if applicable):

"Class II Special Controls Guidance Document: Antimicrobial Susceptibility Test (AST) Systems; Guidance for Industry and FDA"; CLSI M7 (M100-S15) "Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically; Approved Standard"; CLSI M11-A6, "Methods for Antimicrobial Susceptibility Testing of Anaerobic Bacteria; Approved Standard – Sixth Edition."

5. Test Principle:

The Etest® gradient technology is based on a combination of the concepts of dilution and diffusion test methods for susceptibility testing. Etest® directly quantifies antimicrobial susceptibility in terms of discrete MIC values. When the Etest® strip is applied to an inoculated agar plate, the antibiotic is immediately released from the plastic surface into the agar. A predefined, continuous gradient of antibiotic concentrations is created and maintained directly underneath the strip. After incubation whereby bacterial growth becomes visible, a symmetrical inhibition ellipse centered along the strip will be observed. The MIC value in µg/mL is read where the ellipse edge intersects the strip. Since Etest® generates MIC values which fall between two-fold dilutions for interpretation; the MIC value read must be recorded to the next higher two-fold dilution.

6. Performance Characteristics (if/when applicable):

7. Analytical performance:

a. ***Precision/Reproducibility:***

Reproducibility was established using a variety of 26 Gram positive and Gram negative aerobic isolates; 26 *Streptococcus* species; and 26 anaerobes comprised of a variety of *Bacteroides* species, which were evaluated three times at each site, for site to site, and inter site studies. All organisms were tested at all three sites, which demonstrated >95% reproducibility.

b. Linearity/assay reportable range

Not applicable

c. ***Traceability, Stability, Expected values (controls, calibrators, or method):***

FDA recommended Quality Control strains were tested at the concentrations listed (see tables below). The results demonstrated that the device system could produce QC results in the recommended range.

Quality control results demonstrated the ability of the device to produce acceptable results >95% of the time for all QC organisms.

Table 7.c.1. – Tigecycline QC results for aerobic bacteria

Organism	Concentration µg/mL	Reference results	Etest® results
Gram negative			
E. coli ATCC 25922 0.03 – 0.25 µg/mL	<0.032		
	0.032		
	0.064	7	
	0.125	41	55
	0.25	12	2
	>0.25		
Gram positive			
S. aureus ATCC 29213 Expected range 0.032 – 0.25 µg/mL	<0.032		
	0.032		
	0.064	1	2
	0.125	23	40
	0.25	36	18
	0.5		
E. faecalis ATCC 29212 Expected range 0.032 – 0.125 µg/mL	<0.032		
	0.032		
	0.064	17	14
	0.125	43	46
	0.25		
	0.5		

Table 7.c.3. Tigecycline QC Data for *Streptococcus pneumoniae*

Organism	Concentration µg/mL	Reference results	Etest® results
Streptococcus. pneumoniae			
S. pneumoniae ATCC 49619 Expected range 0.016 – 0.125 µg/mL			
	0.016	2	1
	0.032	38	36
	0.064	20	23
	0.125		

Table 7.c.3. – Tigecycline QC results for anaerobic bacteria

Organism	Concentration µg/mL	Reference results	Etest® results
Anaerobes			
Bacteroides fragilis ATCC 25285 Expected range 0.125 - 1 µg/mL	<0.125		
	0.125		
	0.25	31	30
	0.5	29	30
	1		
	>1		
B. thetaiotaomicron ATCC 29741 Expected range 0.5 - 2 µg/mL	<0.25		
	0.25		
	0.5	40	42
	1	20	17
	2		1
	>2		
Eubacterium lentum ATCC 43055 Expected range 0.064 – 0.5 µg/mL	<0.032		
	0.032		
	0.064	35	28
	0.125	25	30
	0.25		2
	0.5		

A 0.5 McFarland is used to determine the correct inoculum.
Colony counts for the clinical / stock isolates were performed periodically at each site to demonstrate that the inoculum procedure results were in the expected CFU/ml range. All colony count inoculum check mean results were within the acceptable ranges.

d. ***Detection limit:***
Not applicable

e. ***Analytical specificity:***
Not applicable

f. ***Assay cut-off:***
Not applicable

2. Comparison studies:
   a. ***Method comparison with predicate device:***
   The reference panels were prepared and tested as recommended by CLSI, including the addition of 2.5 - 5% lysed horse blood when testing *Streptococcus spp.* CLSI recommended broth microdilution and agar dilution

methods were utilized as the reference methods, and were used to compare with the Etest® results. Broth microdilution testing of aerobic organisms for MIC values was performed in testing media that was fresh (<12 hours old).

Clinical testing was performed at three sites. The testing included fresh/stock, clinical aerobic Gram positive and Gram negative isolates; fresh/stock clinical *Streptococcus* isolates; and fresh/stock clinical isolates of a variety of anaerobic strains. Approximately 60% of the Clinical strains were fresh isolates. The Challenge isolates selected for each testing group of organisms were appropriate for this antibiotic; all Challenge strain testing was performed at the sponsor's site. Low numbers from each organism group were tested, although overall the numbers tested were sufficient.

The study included a variety of Gram positive aerobic Clinical isolates and a variety of Gram positive aerobic Challenge isolates, with the following performance (see table below). The FDA provides breakpoints for tigecycline only for *Staphylococcus aureus* (both methicillin-susceptible and methicillin-resistant isolates), and *Enterococcus faecalis* (vancomycin-susceptible isolates only). Therefore, the table below contains performance data for all Gram positive aerobic species for EA and Eval EA, and *S. aureus* and *E. faecalis* only for CA.

Gram positive aerobic isolates

	EA Tot	EA N	EA %	Eval EA Tot	Eval EA N	Eval EA %	CA Tot*	CA N*	CA %	NS
Clinical	**96**	**95**	**99.0**	**96**	**95**	**99.0**	**78**	**78**	**100.0**	**0**
Challenge	**33**	**33**	**100.0**	**33**	**33**	**100.0**	**18**	**18**	**100.0**	**0**
Combined	**129**	**128**	**99.2**	**129**	**128**	**99.2**	**96**	**96**	**100.0**	**0**

***** The CA Total and CA N apply to *S. aureus* and *E. faecalis* only

NS-Non Susceptible
EA-Essential Agreement
CA-Category Agreement
R-resistant isolates
maj-major discrepancies
vmj-very major discrepancies
min- minor discrepancies

Essential agreement (EA) is when the Etest® panels agree with the reference test panel results exactly or within one doubling dilution of the reference method. Category agreement (CA) is when the Etest® panel result interpretation agrees exactly with the reference panel result interpretation. Evaluable (Eval) are results that are within the test range and on scale.

There were 206 Clinical and 30 Challenge isolates from a variety of *Enterobacteriaceae* strains with the following performance.

***Enterobacteriaceae* isolates**

	EA Tot	EA N	EA %	Eval EA Tot	Eval EA N	Eval EA %	CA Tot	CA N	CA %	#R	min	maj	vmj
Clinical	**206**	**205**	**99.5**	**206**	**205**	**99.5**	**206**	**199**	**96.6**	**13**	**7**	**0**	**0**
Challenge	**30**	**30**	**100.0**	**30**	**30**	**100.0**	**30**	**30**	**100.0**	**0**	**0**	**0**	**0**
Combined	**236**	**235**	**99.6**	**236**	**235**	**99.6**	**236**	**229**	**97.0**	**13**	**7**	**0**	**0**

There were 369 *Streptococcus* species, not *S. pneumoniae* isolates tested. Of that total, 298 were Clinical isolates and 71 were Challenge isolates with the following performance.

Streptococcus* species, not *S. pneumoniae

	EA Tot	EA N	EA %	Eval EA Tot	Eval EA N	Eval EA %	CA Tot	CA N	CA %	#R	min	maj	vmj
Clinical	**298**	**298**	**100.0**	**282**	**282**	**100.0**	**282**	**282**	**100.0**	**2**	**0**	**0**	**0**
Challenge	**71**	**71**	**100.0**	**71**	**71**	**100.0**	**71**	**71**	**100.0**	**1**	**0**	**0**	**0**
Combined	**369**	**369**	**100.0**	**353**	**353**	**100.0**	**353**	**353**	**100.0**	**3**	**0**	**0**	**0**

A total of 385 anaerobic bacteria from a variety of *Bacteroides* species, were tested. There were 310 Clinical isolates and 75 Challenge isolates with the following performance.

Anaerobes

	EA Tot	EA N	EA %	Eval EA Tot	Eval EA N	Eval EA %	CA Tot	CA N	CA %	#R	min	maj	vmj
Clinical	**310**	**303**	**97.7**	**310**	**303**	**97.7**	**310**	**300**	**96.8**	**7**	**10**	**0**	**0**
Challenge	**75**	**75**	**100.0**	**75**	**75**	**100.0**	**75**	**73**	**97.3**	**4**	**2**	**0**	**0**
Combined	**385**	**378**	**98.2**	**385**	**378**	**98.2**	**385**	**373**	**96.7**	**11**	**12**	**0**	**0**

The test device had a growth rate of >95%.

i. Matrix comparison:

Not applicable

b. Clinical studies:

i. Clinical sensitivity:

Not applicable

ii. Clinical specificity:

Not applicable

iii. Other clinical supportive data (when a and b are not applicable

Not applicable

c. Clinical cut-off:

Not applicable

d. Expected values/Reference range

Organism	**S**	**I**	**R**
Staphylococcus aureus (including methicillin-resistant isolates)	≤ 0.5	*	*
Streptococcus spp. other than *S. pneumoniae*	≤ 0.25	*	*
Enterococcus faecalis (vancomycin-susceptible isolates only)	≤ 0.25	*	*
Enterobacteriaceae	≤ 2	4	≥8
Anaerobes	≤ 4	8	≥ 16

*The current absence of resistant isolates precludes defining any results other than Susceptible. Isolates yielding MIC results suggestive of "NonSusceptible" category should be submitted to a reference laboratory for further testing.

N. Proposed Labeling:

The expected value range, interpretive criteria and QC are the same as recommended by FDA. All values will be included in the package insert.

O. Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.